

A1  
3' to the carboxy terminus or 5' to the amino terminus of the first sequence, with the flanking sequence having an amino acid sequence substantially corresponding to that found on native HIV I gp41 adjacent said first sequence. In a preferred embodiment of the present invention, the monoclonal antibody ("5-21-3") is described, which is useful as a test reagent in diagnostic assays. In presently preferred forms, body fluid samples from patients are analyzed by immunoassay techniques such as radioimmunoassays, fluorescent immunoassays, or enzyme-linked immunosorbent assays in either direct or competitive formats.

Please replace the paragraph at page 23, lines 23 through 26, with the following replacement paragraph:

A2  
1) Synthesis of the peptide corresponding to gp41 amino acids 121-154 (SEQ ID NO:2).  
NH<sub>2</sub>-Asp-Arg-Glu-Ile-Asn-Asn-Tyr-Thr-Ser-Leu-Ile-His-Ser-Leu-Ile-Glu-Glu-Ser-Gln-Asn-Gln-Gln-Glu-Lys-Asn-Glu-Gln-Glu-Leu-Leu-Glu-Leu-Asp-Lys-COOH

Please replace the paragraph at page 24, starting at line 28 and continuing through page 25, line 11 with the following replacement paragraph:

A3  
The polypeptide was purified by reversed-phase HPLC on C<sub>4</sub> columns, employing gradients of 0.1% TFA/water (A) and 100% acetonitrile (B) as the solvent systems at a flow rate of 1 ml/min for the analytical column (Vydac-214-TP54, Vydac Separation Group, Hesperia, California) or 3 ml/min for the semi-preparative one (Vydac-214-TP510).

The gradient used was:

28% B	1min	28%B	20min	47%B	1min	28%B
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The polypeptide elution from the HPLC column was monitored at 225 nm and 280 nm. The composition of the polypeptide was confirmed by hydrolysis in 6 N hydrochloric acid (HCl)/0.3% phenol at 150°C for 2 hr in vacuo, and subsequently analyzed on a Beckman 6300 amino acid analyzer with a SICA 7000 A integration.

2) Synthesis of the peptide corresponding to gp41 amino acids 126-162 (SEQ ID NO:3).  
NH<sub>2</sub>-Tyr-Asn-Tyr-Thr-Ser-Leu-Ile-His-Ser-Leu-Ile-Glu-Glu-Ser-Gln-Asn-Gln-Gln-Glu-Lys-Asn-Glu-Gln-Glu-Leu-Leu-Glu-Leu-Asp-Lys-Trp-Ala-Asn-Leu-Trp-Asn-Trp-Leu-COOH



Please replace the paragraph at page 27, starting at line 19 and continuing through page 28, line 10 with the following replacement paragraph:

AL  
The stock solution of the peptide thus obtained was reduced with 50 mM DTT at 40°C for 90 minutes. The solution was brought to room temperature, and then dialyzed in a spectrapor membrane (cutoff 6500-8000) against a 0.1 M ammonium acetate buffer, pH 8.1, for 48 hr. The buffer was changed twice. After a total of 72 hr of dialysis, the peptide solution was diluted 3-fold with 0.1 M ammonium acetate buffer, pH 8.1, and allowed to stand in air for 48 hr. A UV spectrum of this peptide solution in water showed a maxima at 276 nm with a shoulder at 289 nm. The peptide was further purified on a reversed-phase C<sub>4</sub> column, and the analyzed as described above, using the following gradient:

30%B   1 min   30%B   20 min   65%B   1 min   30%B

5) Synthesis of peptide corresponding to gp41 amino acids 58-130 (SEQ ID NO:5).

NH<sub>2</sub>-Thr-Val-Trp-Gly-Ile-Lys-Glu-Leu-Gln-Ala-Arg-Ile-Leu-Ala-Val-Glu-Arg-Tyr-Leu-Lys-Asp-Gln-Gln-Leu-Leu-Gly-Ile-Trp-Gly-Cys-Ser-Gly-Lys-Leu-Ile-Cys-Thr-Thr-Ala-Val-Pro-Trp-Asn-Ala-Ser-Trp-Ser-Asn-Lys-Ser-Leu-Glu-Gln-Ile-Trp-Asn-Asn-Met-Thr-Trp-Met-Glu-Trp-Asp-Arg-Glu-Ile-Asn-Asn-Tyr-Thr-Ser-Leu-COOH

6) Synthesis of peptide corresponding to gp41 amino acids 131-175 (SEQ ID NO:6).

NH<sub>2</sub>-Ile-His-Ser-Leu-Ile-Glu-Glu-Ser-Gln-Asn-Gln-Gln-Glu-Lys-Asn-Glu-Gln-Glu-Leu-Leu-Glu-Leu-Asp-Lys-Trp-Ala-Asn-Leu-Trp-Asn-Trp-Leu-Asn-Ile-Thr-Asn-Trp-Leu-Trp-Tyr-Ile-Lys-Leu-Phe-Ile-COOH